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THE COLORABILITY OF GRAPEFRUIT.

By Henry C. Henricksen.

The green rind of a grapefruit contains green coloring matter, chlorophyll and yellow coloring matter, xanthophyll and carotin, which under the microscope appear as minute granules or plastids within the tissue cells. They are therefore called plastid pigments. The tissue also contains other yellow coloring matter which is not in granular form and is therefore called non-plastid pigment. In the color change that takes place before or after the fruit is picked the plastid pigments disappear partly or entirely. The rind of a fruit that is entirely yellow contains no chlorophyll, and it contains much less xanthophyll and carotin than it did when it was green, the yellow color being due primarily to the non-plastid pigments. This shows that the coloring process, whether natural or artificial, involves the destruction of the plastid pigments. This destruction takes place, to some extent, while the fruit is growing, but as long as the growth is vigorous the color of the rind does not change perceptibly because new pigments are formed as fast as old ones are destroyed. When growth ceases, or when it is temporarily arrested, the color of the rind changes because destruction of the pigments continues while formation stops. This explains the reason why the color of fruit changes from green to yellow when the tree becomes dormant and why it changes from yellow to green when the dormancy is broken. It is also a partial explanation of the difference in colorability of different fruit or in fruit from different trees. That can be proved by the difference in catalase content which is usually a good indication of vegetative vigor. There are, however, other contributory factors, for a catalase determination often fails to show that a fruit is or is not readily colorable. Some of those factors are visible, while others are not. Scab is one of the visible factors inhibiting color change, as is also melanose and practically any other rind blemish. Purple scale is especially inhibitive although no other scale, infesting grapefruit in Porto Rico, seems to have any effect on coloring quality. This inhibiting effect is not serious when the fruit is left to color on the tree, but it is when artificial coloring is attempted. In fact a green fruit, the rind of which is scabby or infested with purple scale and which shows a high catalase content, is practically impossible to color properly.

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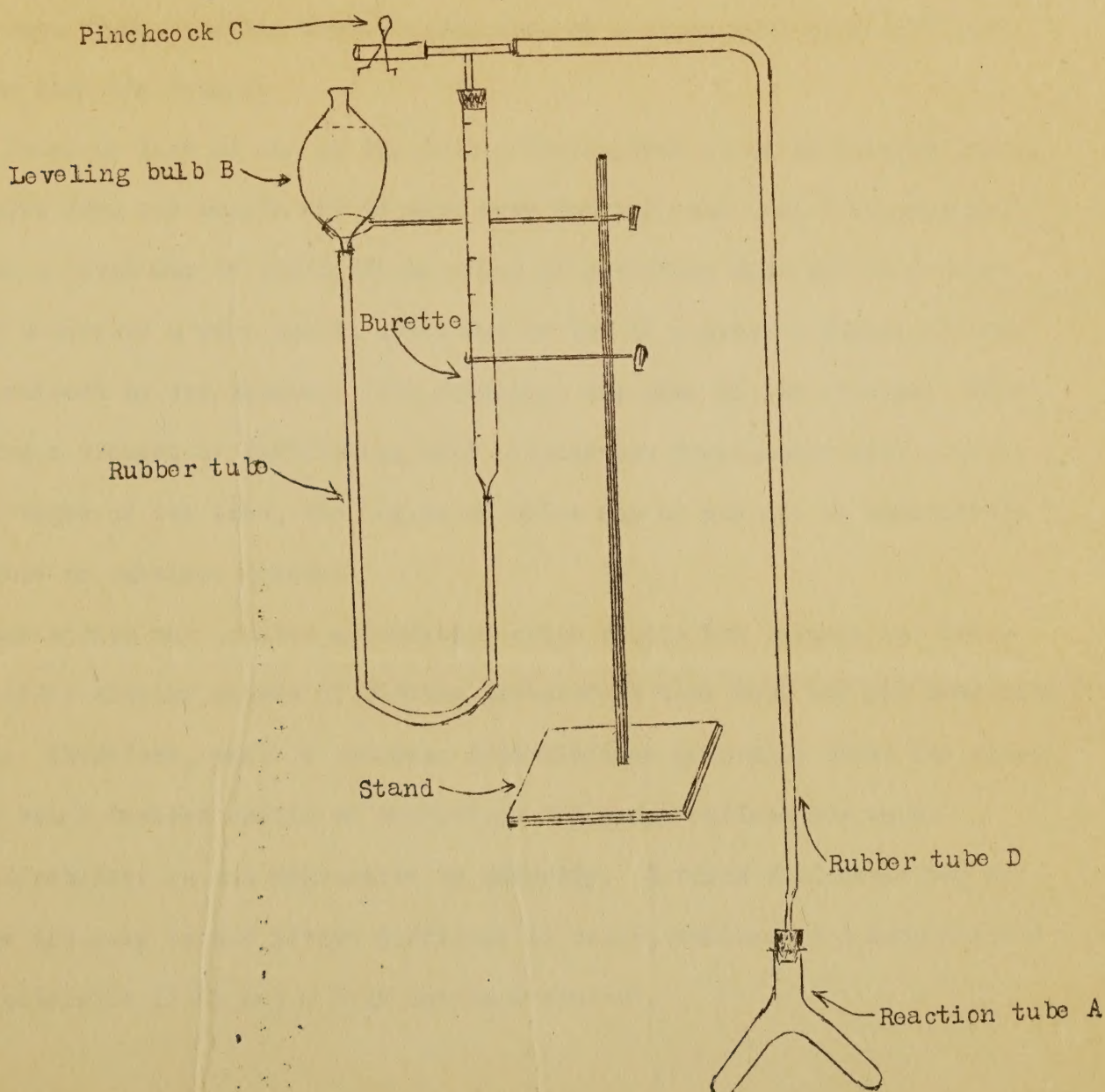
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THE CATALASE TEST. - The following method was used in this investigation: Plugs of rind were removed by means of a 17 millimeter cork borer. Two of these plugs, being 4.54 square centimeters of rind were used. The albedo, which contains but a small amount of catalase, was cut off and only the part containing oil cells was used. That was minced with a sharp knife after which it was mixed with an equal portion of powdered chalk in a small mortar. Water was added to produce a thin pasty consistency after which the mixture was pestled to an impalpable mass. More water was added and the mixture was poured into one prong of the double pronged reaction tube A. The mortar was rinsed with water which was added to the tube together with enough more water to make the total volume 10 cc. Ten cc. of a 3% hydrogen peroxide was measured into the other prong of the tube by means of a pipette. The rubber stopper connected with rubber tube D was then inserted, air tight, in the mouth of the reaction tube, after which the latter was immersed in a dish of tap water. Pinchcock C was opened to let the water level in the burette rise to the zero mark after which it was again closed. In about a minute the temperature of the reaction tube reached equilibrium which was usually 80° to 82° F. since the temperature of the tap water in the laboratory does not vary much during the year. After that the reaction tube was rotated, in the water bath, in such manner that the material and the peroxide became mixed, and the rotation was continued at the rate of one turn every 3 to 4 seconds. At the beginning of the rotation the time was noted on the second dial of a watch and the leveling bulb B was lowered to the 5 cc. mark on the burette. After the water level in the burette dropped to the 5 cc. mark the time was again noted and the bulb lowered to the 10 cc. mark and later to the 20 cc. mark, noting the time the water level reached each mark.



The figures in the following table are representative of those obtained in more than one hundred determinations.

Fruit Nos.	Number of seconds required to liberate oxygen.		
	5 cc.	10 cc.	20 cc.
1	15	30	60
2	17	35	70
3	20	35	65
4	30	60	120
5	25	45	95
6	30	55	110
7	50	100	--
8	45	90	--
9	40	85	--

The Nos. 1, 2 and 3 are typical of fruit the rind of which was found difficult to color. The Nos. 3, 4 and 5 denote fruit the rind of which was found to become well colored in 36 to 48 hours under standard conditions. The Nos. 6, 7 and 8 were found to be readily colorable in 24 to 30 hours under standard conditions.

These conclusions are according to results obtained with fruit from many different plantations during the months of September and October this year, but as stated before there are other governing factors beside the one that is measurable by catalase content. For instance the rind in spots that are bright because of shading contain as much catalase as that of the green areas of the same fruit, but the latter areas are, of course, much more difficult to color than the former.

A catalase test of one or two fruits from a tree is no infallible guide, for fruits from one branch may be much more dormant than that from another. Likewise a fruit may be difficult to color at a certain date and much more readily colorable a week later, which may be due to a greater degree of dormancy produced by dry weather. The condition may also be the reverse. Rain following a drought or fertilizing with nitrogenous fertilizers will produce renewed vigor of the tree, the degree of which may or may not be immediately measurable by catalase content.

Some sprays may produce a condition which upsets the balance and there may be other similar causes of balance disturbance that have not yet been discovered. Therefore, while a catalase determination generally shows the colorability other factors should be considered and proper allowances made.

Colorability is not measurable by maturity. A thick rind enclosing an immature dry pulp is not always difficult to color, neither is a mature fruit easily colorable if it has a high catalase content.

Variation in content of other enzymes, such as peroxidase and reductase, does not furnish a measure of colorability. Neither does variation in pH of the rind tissue. In a green rind the pH is 6, more or less, and in coloring it changes to about 5.

The catalase method may be of interest to dealers and consumers for it furnishes a fairly reliable measure of whether or not the fruit has been artificially colored. Fruit that has been left to color on the tree contains very little catalase, whereas that which has been colored artificially contains as much as it did when picked. That is the catalase is not destroyed by coloring and it persists in the rind of the fruit for at least three to four weeks.

ARTIFICIAL COLORING. - Since ethylene is practically the only product now used in Porto Rico for artificial coloring of citrus fruit this investigation has been confined to the standardization of methods for its use. Such standardization must necessarily be based upon empirical knowledge since neither the chlorophyll change nor the action of ethylene, in the process, is well understood. If it may be admissible to suppose that chlorophyllase or some other enzyme is the chief agency in bringing about the change it may be deduced that ethylene acts as an enzyme accelerator. That its action is such seems reasonable in view of the fact that so small an amount as 1 part by volume to 5000 volumes air is as beneficial as larger amounts.

Unfortunately the small amount does not always produce the desired result and amounts three to four times greater often stimulates the stem-end decay. Standardization of method is therefore very desirable. The first step in that direction is naturally tight coloring rooms. The proper method of operation may be deduced from past and present experience with many different rooms. It is well known that by the use of leaky rooms fruit may often be colored satisfactorily without even loosening the stems or buttons, but the coloring time is much prolonged and if the fruit is difficult to color the proper color may not be attainable under those conditions. This suggests that in order to prevent decay the air or oxygen supply should be kept high, and in order to hasten coloring the ethylene supply should be kept as constant as practicable. It is well known that high temperature accelerates coloring, but it is also well known that it hastens decay. Experience in Porto Rico as well as in Florida shows that under standard conditions satisfactory results can be obtained at 80° F. The coloring rooms should therefore be protected from the direct glare of the sun and while coloring they should never be closed up long enough to allow the temperature to rise perceptibly because of the respiration heat from the fruit.

Before ethylene became known as a coloring agent stove gas was used and with that considerable humidity was thought to be beneficial. Before the advent of stove gas heat together with humidity was used and the process was called sweating. Today the belief in high humidity still persists, but in fact it is neither necessary nor desirable. With ethylene the coloring proceeds well with the natural humidity of the air in Porto Rico together with that given off by the fruit. It needs not be feared that the loss of moisture from the fruit will cause excessive shrinkage, for it will not be in the coloring room long enough for that. The removal of some moisture from the rind is desirable as it helps to toughen it. Also it is desirable to keep the air surrounding the fruit as dry as possible for the sake of preventing decay.

On the basis of general coloring room experience here and in Florida coupled with results obtained from small scale experiments where all the conditions were controlled the following method is recommended: Have the rooms tight and fitted with suitable blowers in such manner that the air can be forced through the fruit in the stacked boxes rather than over or between the boxes. Apply ethylene gas as follows: 1 cubic foot to 3000 cubic feet room space. Two hours later repeat the charge. Two hours after that ventilate the room for 5 to 15 minutes according to the efficiency of the ventilating system, after which apply gas at the rate of 1 to 5000. Repeat the charge of 1 to 5000 at about two hours intervals, with a ventilation period following each gassing period, until the fruit is colored. If a coloring room is not very tight the gas charge may be increased slightly and the gas may be left in the room for a longer period. It seems best, however, to make the room fairly tight and to keep the gassing periods below three hours.

